Territorial response of Carolina chickadee (*Poecile carolinensis*) to playback of Carolina chickadee and black-capped chickadee (*Poecile atricapillus)* song

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Abstract

Two morphologically similar sister species, Carolina chickadee (Poecile carolinensis) and black-capped chickadee (Poecile atricapillus), distinguished primarily by song, have distinct geographic ranges except for a thin band of overlap. I used a play-back experiment to determine if Carolina chickadees perceive black-capped chickadees as conspecific or heterospecific by investigating differences in territorial responses to song playback in Columbus, Ohio, approximately 55 miles south of the hybrid zone in northern Ohio. I hypothesized that Carolina chickadees would show a significant difference in territorial response to conspecific song than to black-capped chickadee song. Seven Carolina chickadees were presented with both a Carolina and black-capped chickadee song. I recorded the number of songs sung by territorial males during pre-playback, playback, and post-playback and the minimum approach distance to the speaker. The results showed that in general the Carolina chickadees responded more aggressively to Carolina song, however the differences did not quite reach significance ($p=0.063$). The results show that in a historically Carolina chickadee range, chickadees discriminate between species, suggesting that the sympatry between the two species is evolutionarily recent since no discrimination is observed within the hybrid zone.

Introduction

Carolina chickadees (*Poecile carolinensis*) and black-capped chickadees (*Poecile atricapillus*) are small, morphologically similar sister species of the family Paridae (chickadees and titmice) with differing habitats and distributions (Mostrom et al. 2002; Foote et al. 2010). Carolina chickadees inhabit deciduous forests of the southeastern United States (Mostrom et al. 2002), while black-capped chickadees are found in the mixed deciduous/coniferous woodlands of the northern United States and Canada (Foote et al. 2010). Carolina chickadees are found from the east coast west to Illinois and Kansas and south to central Florida with a northern boundary in northern Ohio and Indiana (Mostrom et al. 2002). Black-capped chickadees range from coast to coast with a southern boundary in central Kansas, central Missouri, and central Indiana and a northern boundary in western Alaska and north-central Ontario (Foote et al. 2010). There are areas in the United States where the two sibling species have overlapping ranges and interbreed . This hybrid zone occurs in a narrow band across the eastern United States including Pennsylvania, west Maryland, West Virginia, Virginia, and northern Ohio (Mostrom et al. 2002) (Figure 1). The band is becoming wider and expanding northward (Reudink et al. 2007). The northward expansion and shift of the hybrid zone suggests that Carolina chickadees have an advantage over black-capped chickadees (Reudink et al. 2007). Bronson et al. (2003) found that Carolina chickadee males are dominant over black-capped chickadee males and that females prefer to pair with dominant males, which in the wild results in females preferring Carolina males over black-capped chickadee males and may be contributing to the northward expansion of the hybrid zone.

While black-capped and Carolina chickadees are morphologically very similar, they are genetically distinct (Mostrom et al. 2002; Foote et al. 2010). However, there are a few subtle

morphological differences between the two species: black-capped chickadees have more white on the outer edges and tips of the greater wing-covers and edges of the outer secondaries and are generally larger in size (Mostrom et al. 2002; Foote et al. 2010).

Black-capped and Carolina chickadees also differ in their vocalizations. The most common song type ("Type A" song) of Carolina chickadees consists of a *high-low-high-low* pattern of whistled notes that usually occurs in 4-note sets (Ward 1966, Mostrom et al. 2002). The number of notes in the song can vary across the species' range and between individuals (Ward 1966). One song type of Black-capped chickadees consists of a 2 note pattern of *fee-bee* where the first note is higher than the second (Foote et al. 2010). The song is not variable across the majority of the species' range (Kroodsma et al. 1999). Individuals may sing aberrant songs in the black-capped/Carolina chickadee hybrid zone (Enstrom and Bollinger 2009), but many hybrids include perfect versions of songs from both species in their repertoire (Brewer 1963, Wright 2010).

Since black-capped and Carolina chickadees can hybridize, and are morphologically similar, it is important to consider whether Carolina chickadee males differentiate conspecifics from black-capped chickadees. Observing the intensity of aggressive behavior in a species can give insight into a possible discrimination of conspecifics from heterospecifics since individuals tend to be more aggressive toward conspecifics than heterospecifics (Emlen et al. 1975). It can also give insight into the recency of sympatry between two closely related species. Often in areas of overlap between congeners, the two species evolve character displacement and show inter-specific discrimination (Lynch and Baker 1991). However if the sympatry is recent, often congeners experience intense competition and do not show discrimination. Studies have shown that there are no differential responses to the two chickadee species' songs within the hybrid

zone (Ratcliffe and Weisman 1986, Wright 2010). Furthermore, Brewer (1963) observed that Carolina chickadees use aggression to defend their territories from black-capped chickadees as well as hybrid chickadees in the hybrid zone. This aggression shows that there is intense competition for territories, mates and resources (Foote et al. 2010).

The objective of this study is to determine whether Carolina chickadees treat black-capped chickadees as conspecific or heterospecific in territorial interactions in an area where Carolina chickadees do not regularly compete with black-capped chickadees. To test this question, I observed whether wild, territorial male Carolina chickadee males responded differentially to Carolina chickadee song than to black-capped chickadee song. Specifically, I examined the difference in number of songs elicited during and after playback and the nearest approach distance to the speaker. If Carolina chickadees perceive black-capped chickadees as a conspecific (and therefore compete for a mate and other resources) then Carolina chickadee males will react to the black-capped song in a manner similar to their own species' song. However, if Carolina chickadees respond differently to the two songs then I assumed that they perceive one song as heterospecific. I hypothesize that Carolina chickadee males will show a differential response, responding more weakly to black-capped chickadee songs than to the conspecific songs, since the Carolina chickadees tested are not within the hybrid zone and do not consistently interact with black-capped chickadees.

Methods

Study Area

I conducted my study in Columbus, Ohio, during May 2012 at various locations in and directly surrounding the Ohio State University main campus. Columbus, Ohio, is approximately 55 miles south of the current hybrid zone in northern Ohio. During the breeding season, only

Carolina chickadees are present, however a few black-capped chickadees are reported during the winter months in various places around Columbus (eBird 2012). I used the following locations: West Campus Woodlot, Olentangy River Wetland Research Park, Chadwick Lake, Tuttle Park, and outside of the Agriculture Administration building (Figure 2). All of the sites were small wooded areas in a larger urban matrix.

Selection of Individual

I used 10 unmarked, wild, territorial male Carolina chickadees as subjects. I determined the territories of the chickadees by mapping their singing post during the first three hours after dawn. I followed each male and observed him for at least 30 minutes to determine the extent of each bird's territory since chickadees use song to mark and defend territories (Brewer 1961). Males were distinguished from females by the presence of singing, since only males sing the "Type A song" (Mostrom et al. 2002). I used the estimated territory boundaries to individually identify chickadees. I sampled birds whose territories were at least 150 meters apart.

Playback Experiment

I created 20 playback files (10 of each species) using 20 different recordings that contained 3 distinct playback periods which were 5 minutes each: a passive pre-playback period, an active playback period, and a passive post-playback period (Wright 2010). I obtained the recordings from the Borror Laboratory of Bioacoustics at the Museum of Biological Diversity digital sound database, ensuring the recordings were not from neighboring males. During the active playback period, the songs were broadcast at a rate of 10 songs per minute, which is the average rate of song for Carolina chickadees (Ratcliffe and Weisman 1992).

I conducted the playback experiments from May 5 to May 20, 2012, during the first 3 hours following sunrise. I mounted a player and speaker on a tripod at a height of 1 meter from

the ground in the center of each territory. After waiting 1 minute to allow the bird to settle down, I broadcasted the playback file, recording vocalizations for the three 5-minute periods, and, in 1 meter increments, recording the chickadee's distance from the speaker every 30 seconds just during the active playback period. All chickadees were more than 10 meters away from the speaker before the active playback period began. I recorded vocalizations using a Marantz PMD 670 solid-state recorder (Marantz Professional, Kanagawa, Japan) and Audio-Technica AT815 "shotgun" microphone with Rycote windscreens (Rycote Ltd, Stroud, U.K.). To determine which playback species to play during the first playback of each territory, I used a random number generator. Once a species was randomly generated, I assigned each playback file a number and used the random number generator to select which playback file to play for each territory. I played the other species' song within two days of the first playback at the same territory to reduce the chance of territory turnover and ensure the identity of the individual. Once a song file was played, I did not use it again throughout the duration of the experiment, to eliminate the possibility of pseudoreplication. I did not conduct playbacks on neighboring territories on the same day and only conducted 1 playback on each territory in a single day.

Statistical Analysis

I analyzed the difference in total number of songs elicited during the playback period and the pre-playback period and the difference in total number of songs elicited during the post-playback period and pre-playback period (Table 1) to adjust for the song rates of the individual birds. I also analyzed the nearest approach distance to the speaker during the active playback period. For each of these variables I used the Wilcoxon paired signed-rank test to determine significance. I used the exact p-value with an alpha level of 0.05.

Results

My sample size was 7 male Carolina chickadees, since at 3 out of the 10 territories I identified, the males did not respond or come near the playback experiment. This could be due to territory change, mortality of the bird, or the bird was off territory when I was conducting the experiment.

The median difference of total songs sung during the playback period was 0 with a range from -4 to 27 for the black-capped chickadee playback (BCCH) and 16 with a range from 0 to 61 for the Carolina chickadee playback (CACH) (Figure 3). The median difference of total songs sung during the post-playback period was 0 with a range from -4 to 0 for the BCCH stimulus and 4 with a range from 0 to 47 for the CACH stimulus (Figure 4). The median minimum approach distance was 3 for the BCCH playback with a range from 0 to 10 and 0 for the CACH playback with a range from 0 to 1 (Figure 5). The range for the minimum approach distance for the BCCH playback was 0 to 10 meters while the range was 0 to 1 meters for the CACH playback (Figure 10). Males tended to sing more during the playback period in response to CACH than to BCCH (Wilcoxon signed-ranks, $P = 0.156$) Males tended to sing more during the post-playback period in response to CACH than to BCCH (Wilcoxon signed-ranks, $P=0.063$). Males also tended to have a lower minimum approach distance to CACH than to BCCH (Wilcoxon signed-ranks, $P=0.063$).

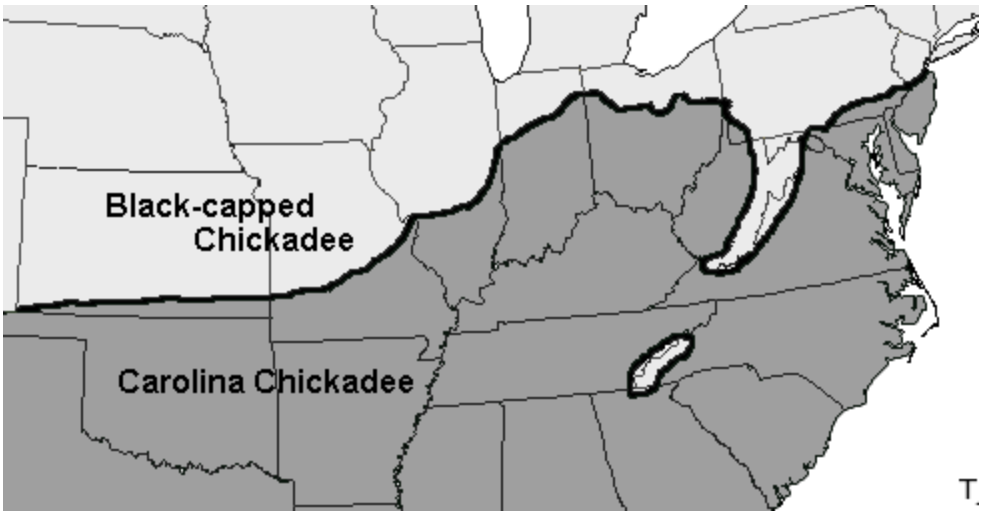

Figure 1. Range map of black-capped chickadee and Carolina chickadee ranges.

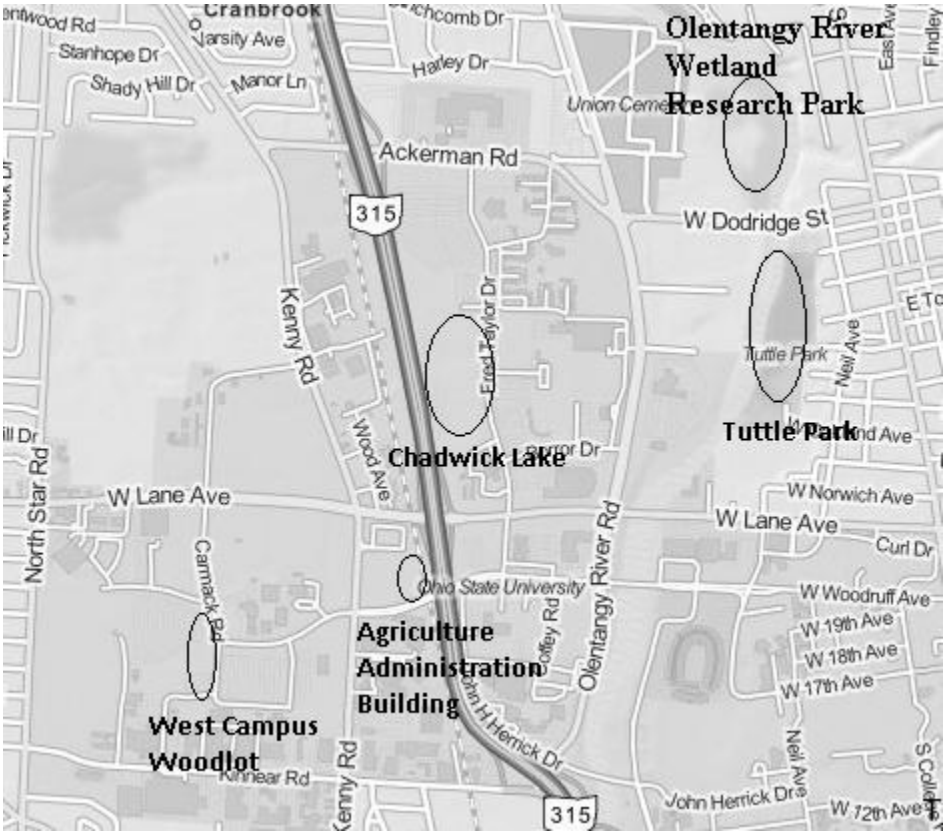

Figure 2. Map of study area in Columbus, Ohio.

Bird ID	Playback Species	Pre-Playback	Playback	Post-Playback	Difference between Playback and Pre-Playback	Difference between Post- and Pre-Playback
1	BCCH	4	0	0	-4	-4
1	CACH	0	18	13	18	13
2	BCCH	0	0	0	0	0
2	CACH	0	12	1	12	1
3	BCCH	0	0	0	0	0
3	CACH	0	31	9	31	9
4	BCCH	0	0	0	0	0
4	CACH	0	0	0	0	0
5	BCCH	0	10	0	10	0
5	CACH	0	16	4	16	4
6	BCCH	1	28	1	27	0
6	CACH	0	14	0	14	0
7	BCCH	0	9	0	9	0
7	CACH	0	61	47	61	47

Table 1. Number of songs elicited during pre-playback, playback and post-playback periods for each territory and each playback species (BCCH and CACH) in Columbus, Ohio, during May 2012.

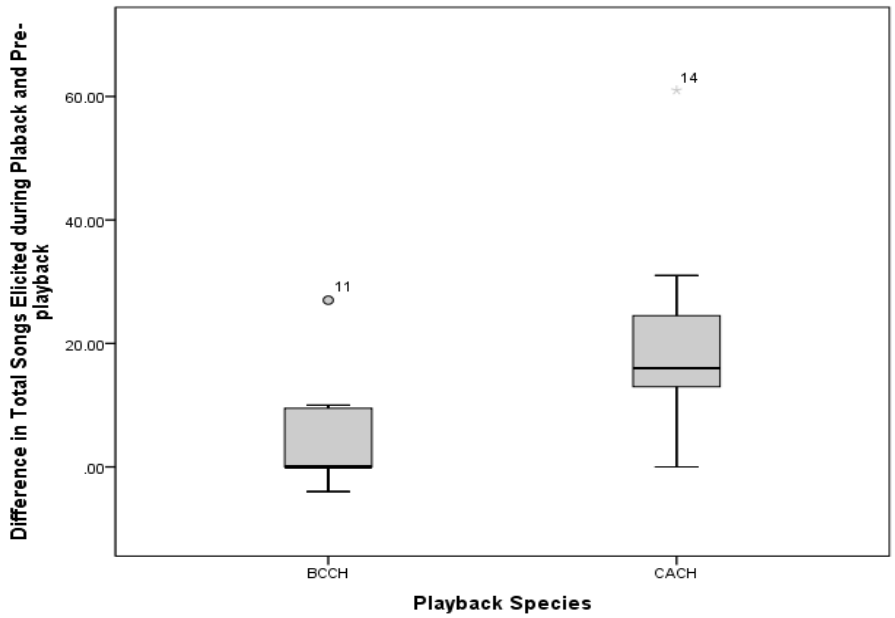

Figure 3. Box-plot of difference in total songs elicited during playback and pre-playback in for each playback species in Columbus, Ohio, during May 2012.

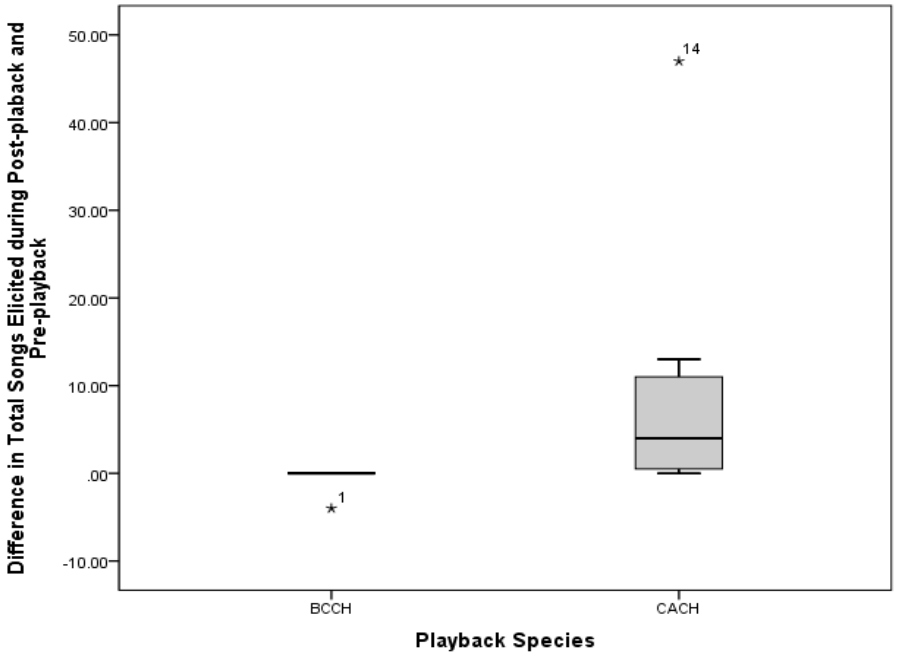

Figure 4. Box-plot of differences in total songs elicited during post-playback and pre-playback periods for each playback species in Columbus, Ohio, during May 2012.

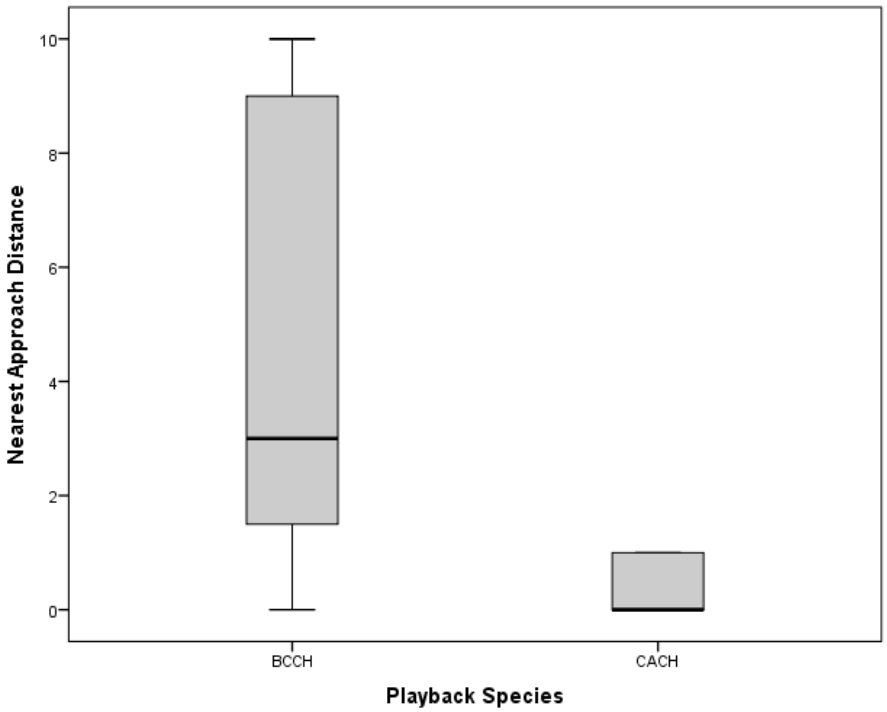

Figure 5. Box-plot of nearest approach distance for each playback species in Columbus, Ohio, during May 2012.

Discussion

Differences in responses to song playbacks of the two different species were not statically significant due to small sample size. Nevertheless Carolina chickadees showed a clear differential aggressive response to CACH playback than to BCCH playback, singing 16 times more often during the playback period and approaching 3 times closer to the speaker. My results were consistent with findings from other studies. Kershner (1999) found that while there was no differential territorial response in areas in the center of the hybrid zone, the amount of differential response increased with distance from the center of the hybrid zone. Since my study area was approximately 55 miles south of the current hybrid zone, a clear differential response matches with Kershner's findings. These finding suggest that Carolina chickadees discriminate between conspecifics and heterospecifics in areas where there is no competition for breeding sites and mates.

Since other studies have found that chickadees do not differentiate conspecifics from heterospecifics (Kershner 1999, Ratcliffe and Weisman 1986, Wright 2010), my results also supported the idea that the hybrid zone between the two chickadee species is recent on an evolutionary time scale. Character displacement develops to reduce competition for territories, mates, and resources in areas where 2 closely related species have co-existed for a long time. Individuals within the sympatric area show greater disparity in certain characteristics, such as beak size or vocalizations, than individuals outside of the sympatric area. Individuals within the hybrid zone typically show discrimination between species if character displacement has occurred (Lynch and Baker 1991). There are two plausible reasons for the lack of species discrimination in sympatric areas,. The first is that character displacement has not occurred, causing intense competition between the two species. Not discriminating between the two

species thus increases the likelihood of securing a territory, defending a territory, and protecting a mate. The second is that the species cannot discriminate between the two species, regardless of the level of competition. My results showed that Carolina chickadees were able to discriminate between calls of the two species in the absence of competition so the recency of sympatry and lack of character displacement are more plausible hypotheses.

Further research could determine if the findings that chickadees show differential response in allopatric areas holds true for areas north of the hybrid zone in historically black-capped chickadee range. They could examine the question if black-capped chickadees will discriminate in areas where they do not compete with Carolina chickadees even though Carolina chickadees have been found to be dominant to black-capped chickadees in the hybrid zone (Bronson et al. 2003). Further research could also examine if character displacement is beginning to develop within the hybrid zone, by examining if there is more variation in morphological features, vocalizations, or behaviors within the hybrid zone than outside of it. Character displacement could reduce the amount of hybridization and the introgression of Carolina chickadee genes into the less-dominant black-capped chickadee genes and the reduction of the black-capped chickadee range.

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